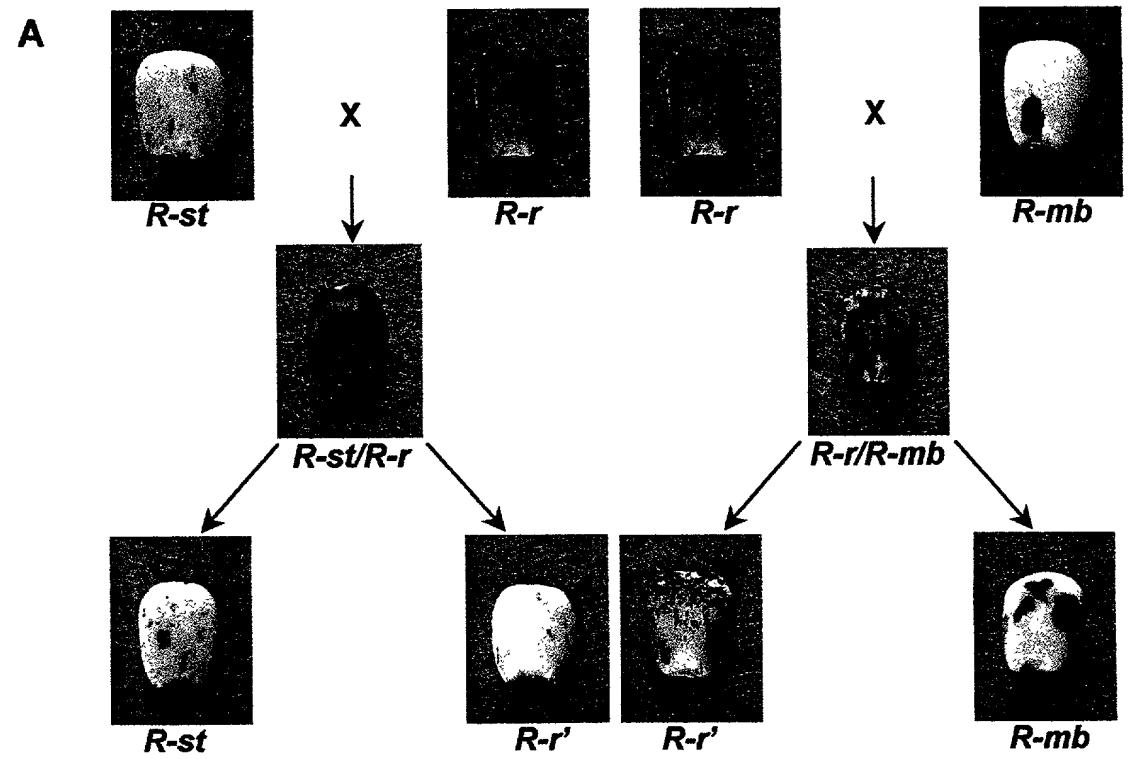
ABSTRACT OF THE DISCLOSURE

[00507] Transgenic silencing is a little understood process by which genes introduced into plants are turned off or silenced. Genetic screens were designed to identify corn mutants with reduced gene silencing activity. Such mutant corn lines include *Mop1-1*; *Mop1-2EMS*; *Mop2-1*, *mop3-1*; *CC2343*, *rmr1-1*; *rmr1-2*; *rmr2-1*; *rmr6-1*; *rmr7-1*; *rmr7-2*; *rmr8-1*; *rmr9-1*; *Mop1-4*; *Mop1-5*; and *rmr11-1* and seeds derived therefrom, the plants are useful for corn breeding programs to produce inbred and hybrid seed with reduced gene silencing activity.


A
X
X
R-st
R-r
R-r
R-mb
R-st/R-r
R-r/R-mb
R-st
R-r'
R-r'
R-mb

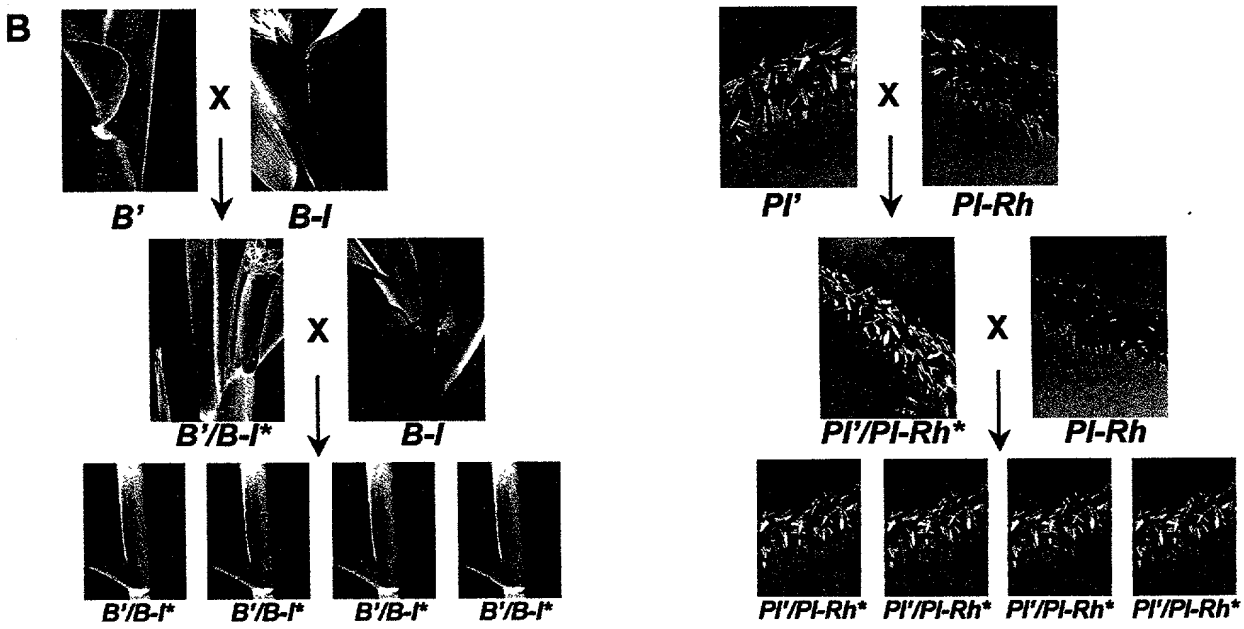
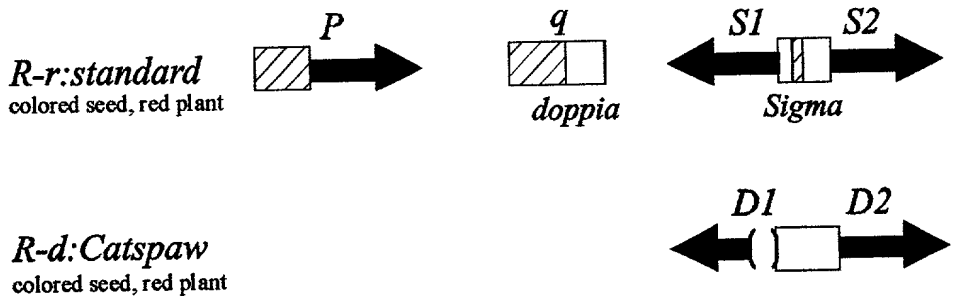
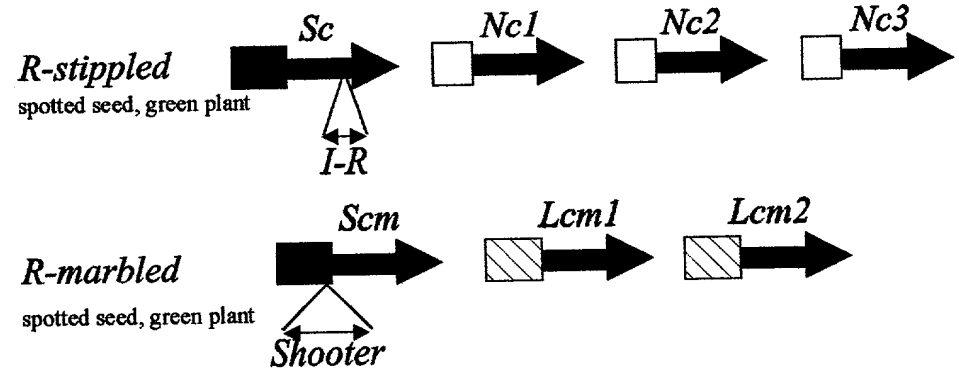

B
X
X
B'
B-l
Pl'
Pl-Rh
B'/B-l*
B-l
Pl'/Pl-Rh*
Pl-Rh
B'/B-l*
B'/B-l*
B'/B-l*
B'/B-l*
Pl'/Pl-Rh*
Pl'/Pl-Rh*
Pl'/Pl-Rh*
Pl'/Pl-Rh*

Figure 1

A **Paramutable**



Paramutagenic



B

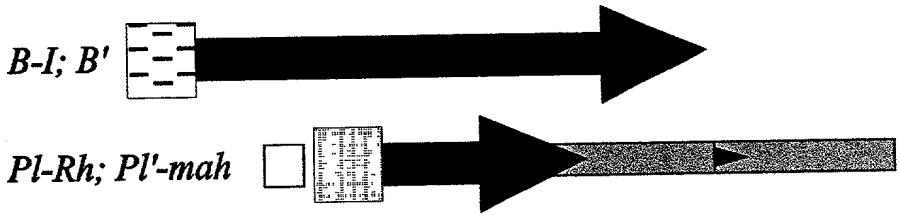


Figure 2

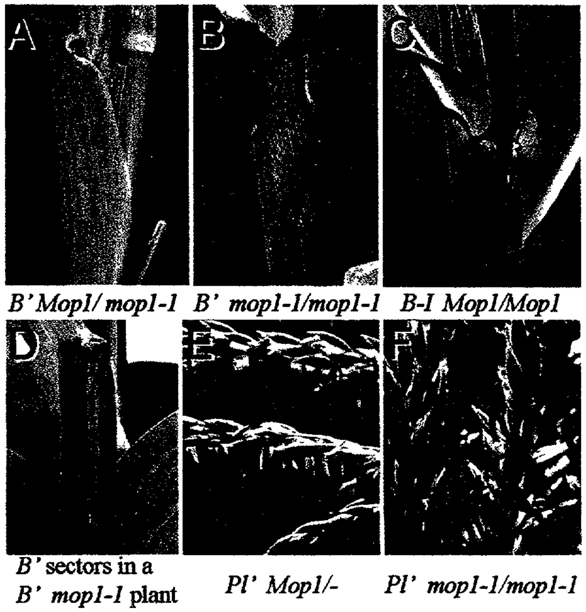

A
B
C
B' Mopl/ mopl-1
B' mopl-1/mopl-1
B-I Mopl/Mopl
D
E
F
B' sectors in a *B' mopl-1* plant
Pl' Mopl/-
Pl' mopl-1/mopl-1

Figure 3

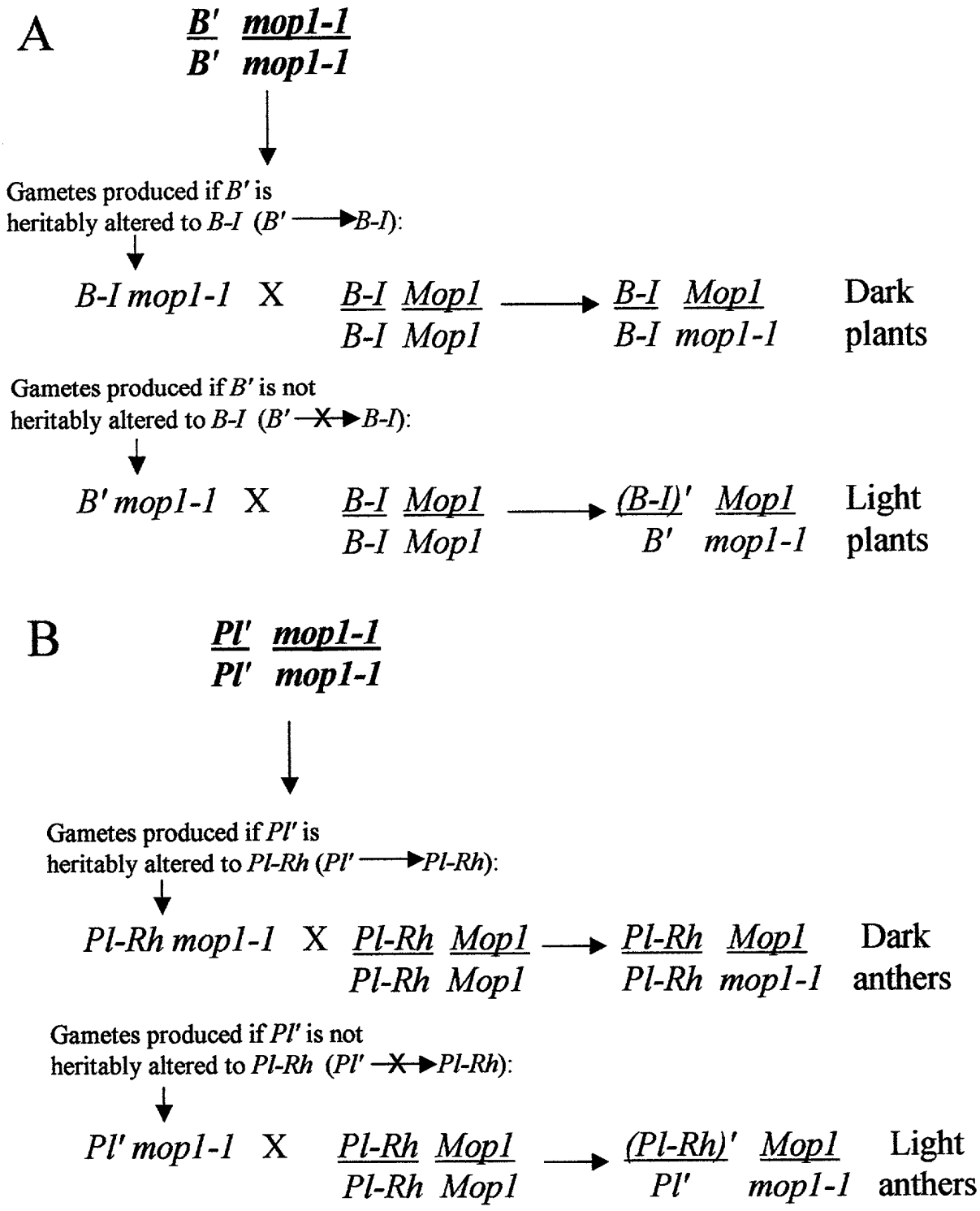

A
B' mop1-1
B' mop1-1
Gametes produced if B' is heritably altered to B-I (B' → B-I):
B-I mop1-1 X B-I Mop1 / B-I Mop1 → B-I Mop1 / B-I mop1-1 Dark plants
Gametes produced if B' is not heritably altered to B-I (B' → X B-I):
B' mop1-1 X B-I Mop1 / B-I Mop1 → (B-I)' Mop1 / B' mop1-1 Light plants
B
Pl' mop1-1
Pl' mop1-1
Gametes produced if Pl' is heritably altered to Pl-Rh (Pl' → Pl-Rh):
Pl-Rh mop1-1 X Pl-Rh Mop1 / Pl-Rh Mop1 → Pl-Rh Mop1 / Pl-Rh mop1-1 Dark anthers
Gametes produced if Pl' is not heritably altered to Pl-Rh (Pl' → X Pl-Rh):
Pl' mop1-1 X Pl-Rh Mop1 / Pl-Rh Mop1 → (Pl-Rh)' Mop1 / Pl' mop1-1 Light anthers

Figure 4

Stability of *Pl'* in *mop1-1/mop1-1* vs. wild-type sibs

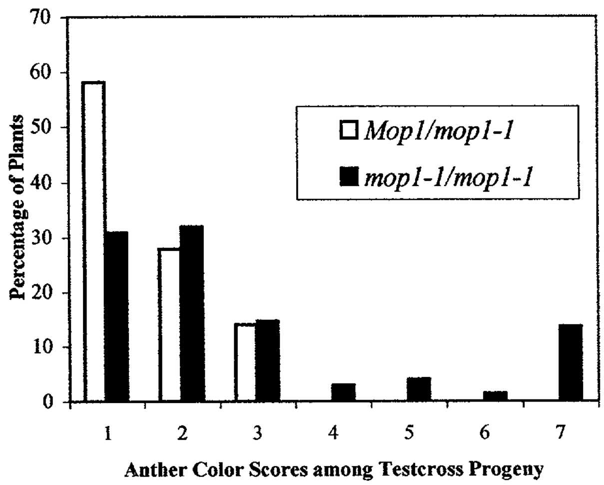


Figure 5

A

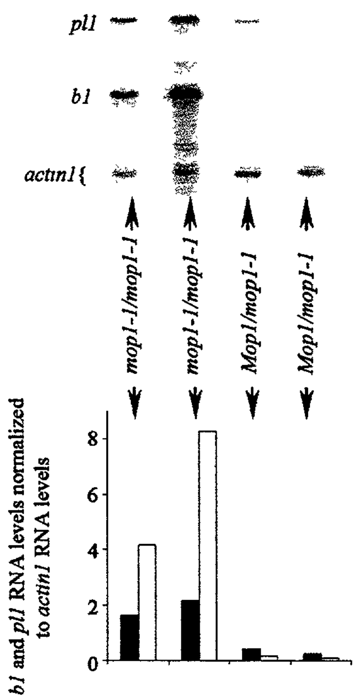


Figure 6

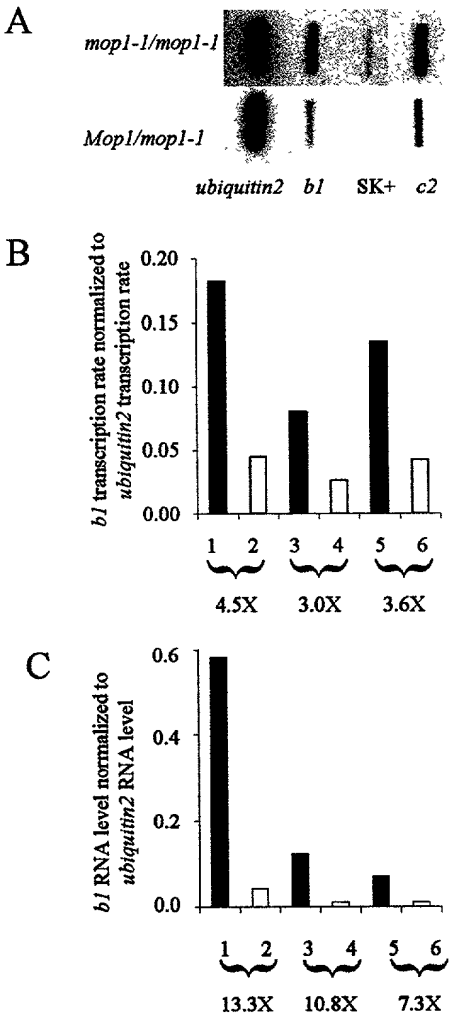

A
mop1-1/mop1-1
Mop1/mop1-1
ubiquitin2
b1
SK+
c2
B
b1 transcription rate normalized to ubiquitin2 transcription rate
0.20
0.15
0.10
0.05
0.00
1 2 3 4 5 6
4.5X
3.0X
3.6X
C
b1 RNA level normalized to ubiquitin2 RNA level
0.6
0.4
0.2
0.0
1 2 3 4 5 6
13.3X
10.8X
7.3X

Figure 7

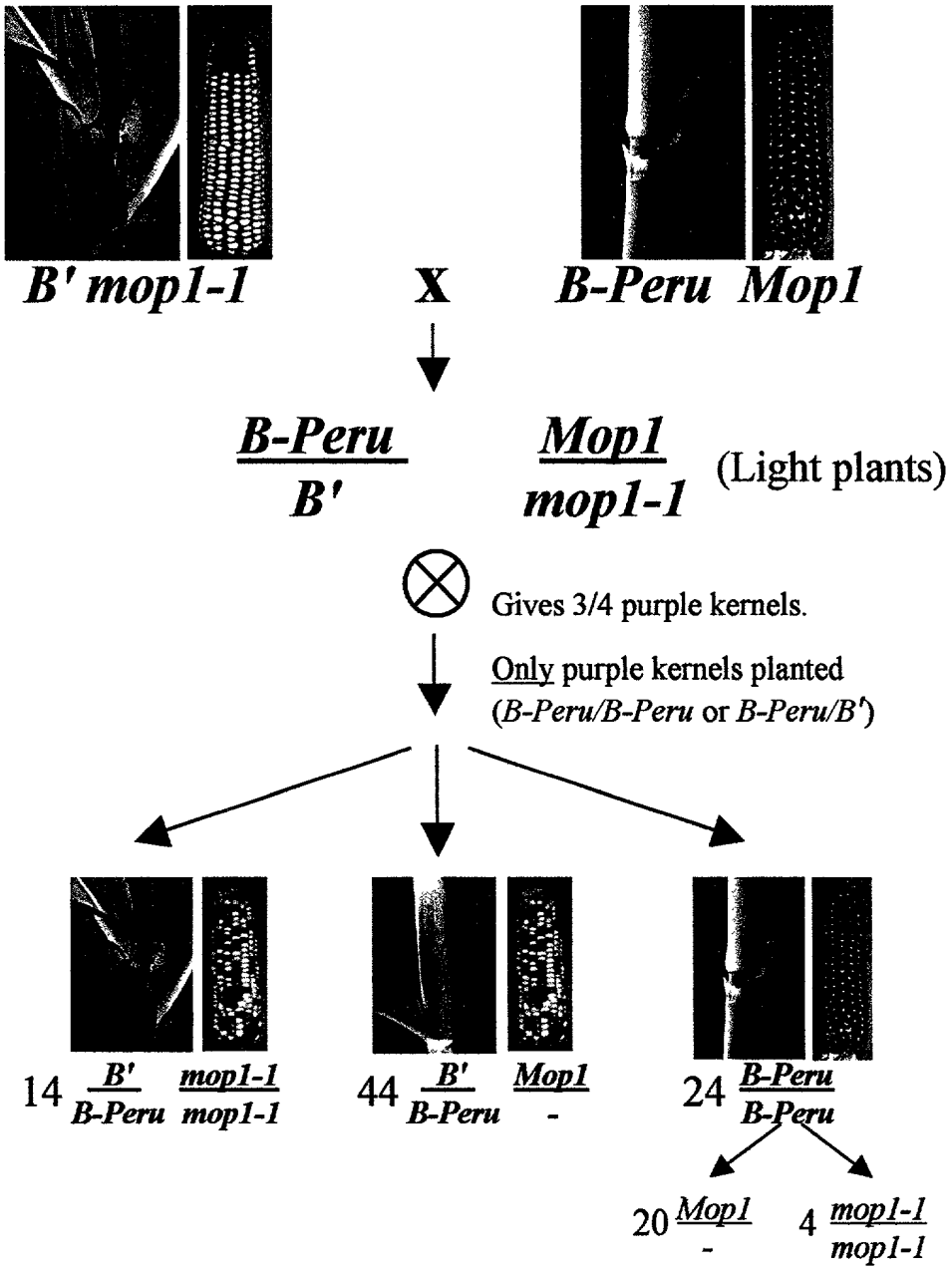
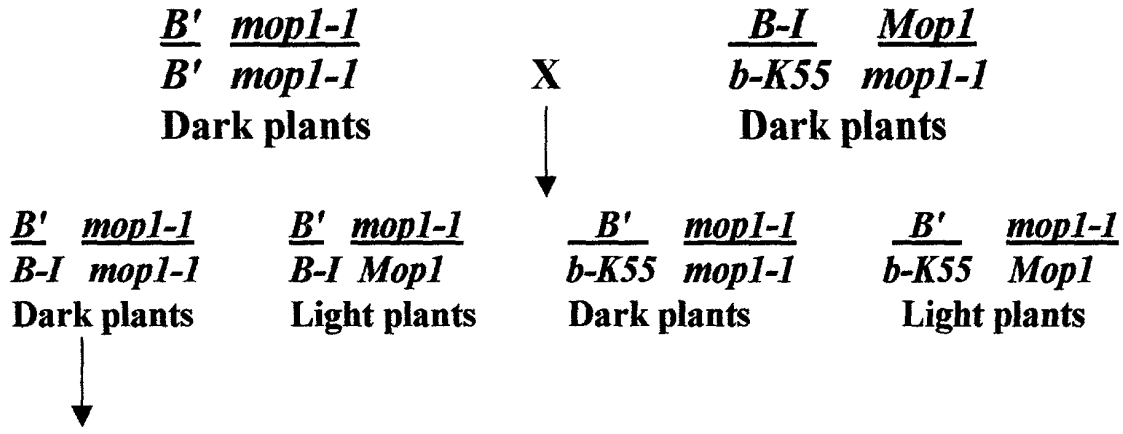

B' mop1-1
X
B-Peru Mop1
B-Peru / B'
Mop1 / mop1-1
(Light plants)
Gives 3/4 purple kernels.
Only purple kernels planted
(B-Peru/B-Peru or B-Peru/B')
14 B' / B-Peru mop1-1 / mop1-1
44 B' / B-Peru Mop1 / -
24 B-Peru / B-Peru
20 Mop1 / -
4 mop1-1 / mop1-1

Figure 8



Testcross (by *B-Peru/B-I Mop1/Mop1* tester--diagram shows only *B-Peru* gametes of tester)

Gametes produced if paramutation is prevented (*B-I* —X→ *B'*):

50% B' mop1-1 X *B-Peru Mop1* ⟶ *B' mop1-1* / *B-Peru Mop1* Light plants

50% B-I mop1-1 X *B-Peru Mop1* ⟶ *B-I mop1-1* / *B-Peru Mop1* Dark plants

Gametes produced if paramutation occurred (*B-I*⟶*B'*):

100% B' mop1-1 X *B-Peru Mop1* ⟶ *B' mop1-1* / *B-Peru Mop1* Light plants

Figure 9

A

R-st	***B-bar***	***pl-sr***	***Mop1***			***R-d***	***B-bar***	***pl-sr***	***Mop1***
r-g	***B'***	***Pl'***	***mop1-1***	**X**		***r-g***	***B'***	***Pl'***	***mop1-1***

Four genotypes of interest among *R-d/- Pl'/-* progeny:

3	***R-d***	***Mop1***	Light plant
16	***r-g***	**-**	
3	***R-d***	***Mop1***	Light plant
16	***R-st***	**-**	
1	***R-d***	***mop1-1***	Dark plant
16	***r-g***	***mop1-1***	
1	***R-d***	***mop1-1***	Dark plant
16	***R-st***	***mop1-1***	

B

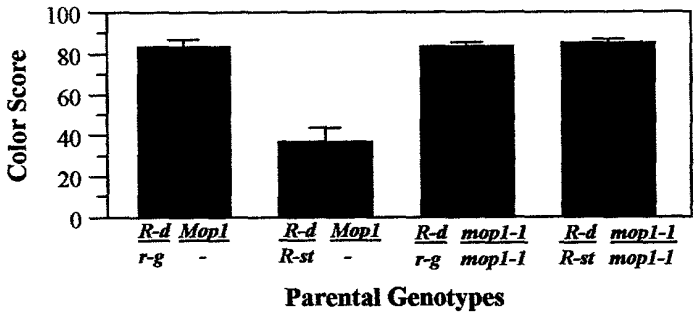


Figure 10

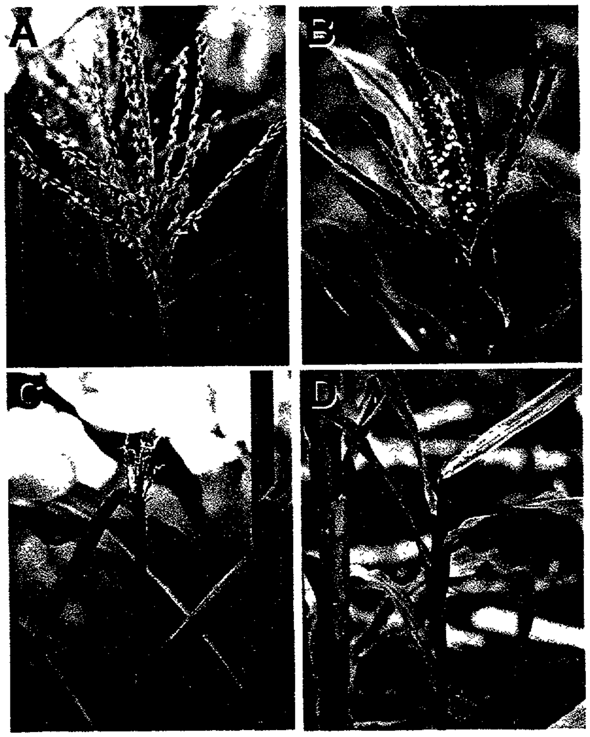


Figure 11

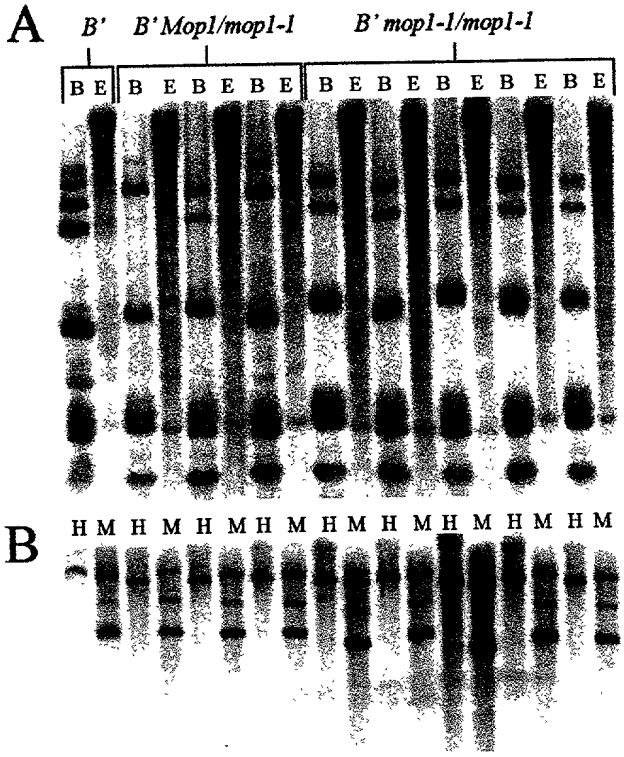

A
B'
B' Mop1/mop1-1
B' mop1-1/mop1-1
B E B E B E B E B E B E B E B E B E B E
B
H M H M H M H M H M H M H M H M H M H M

Figure 12

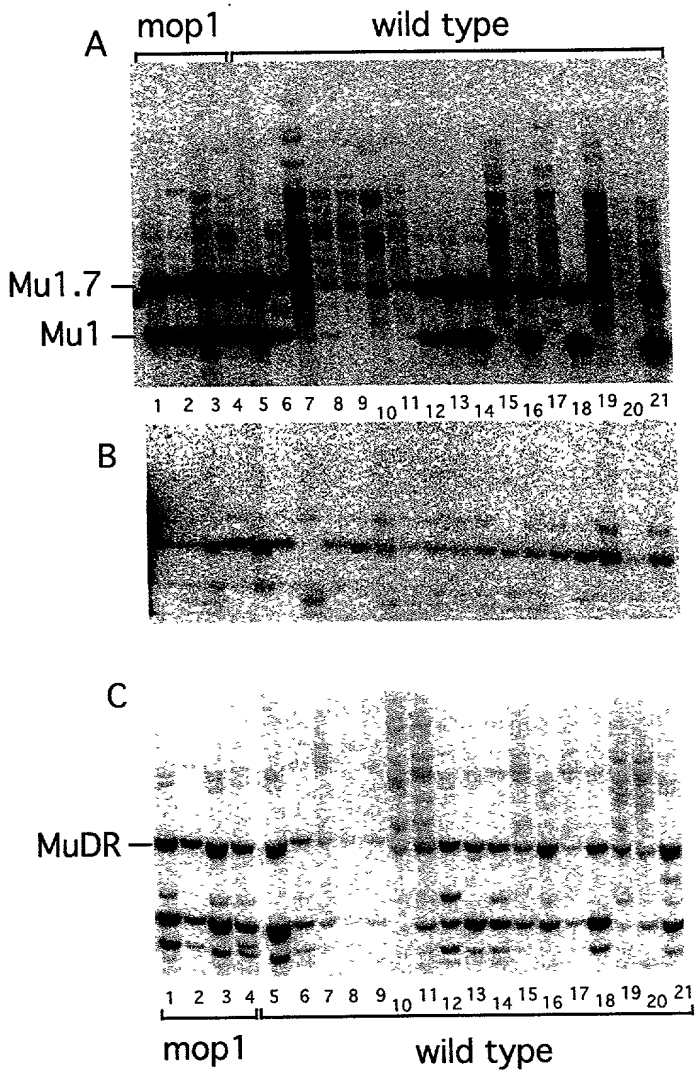

A
mop1
wild type
Mu1.7
Mu1
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21
B
C
MuDR
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21
mop1
wild type

Figure 13

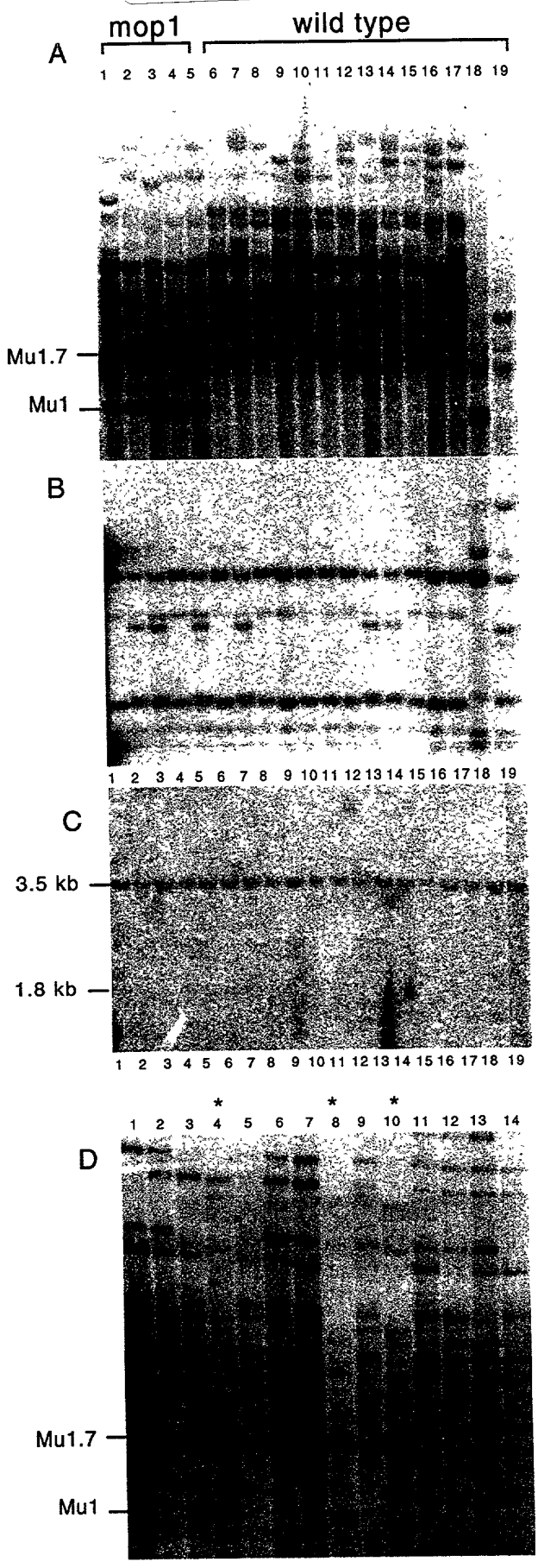

mop1
wild type
A
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Mu1.7
Mu1
B
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
C
3.5 kb
1.8 kb
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
D
1 2 3 4 5 6 7 8 9 10 11 12 13 14
Mu1.7
Mu1

Figure 14

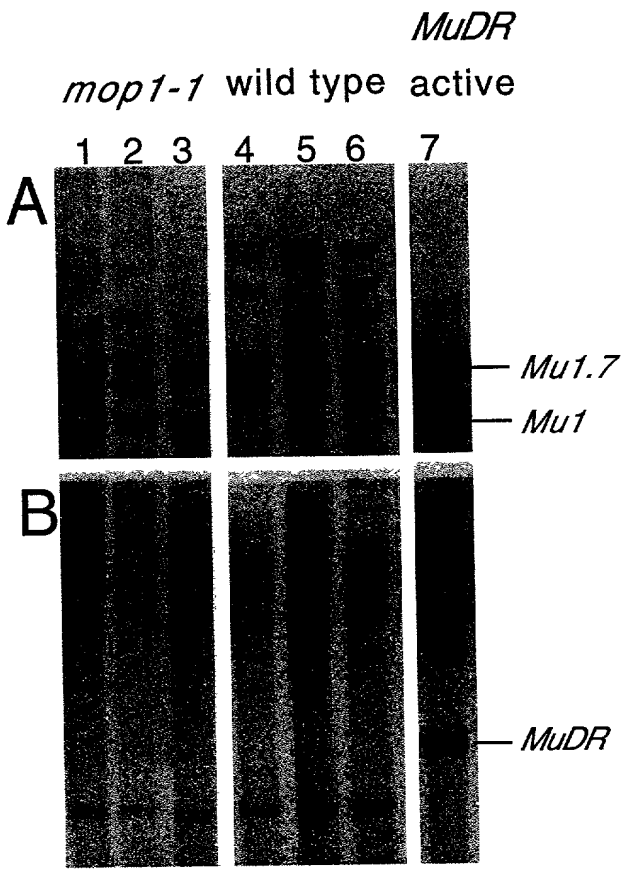


Figure 15

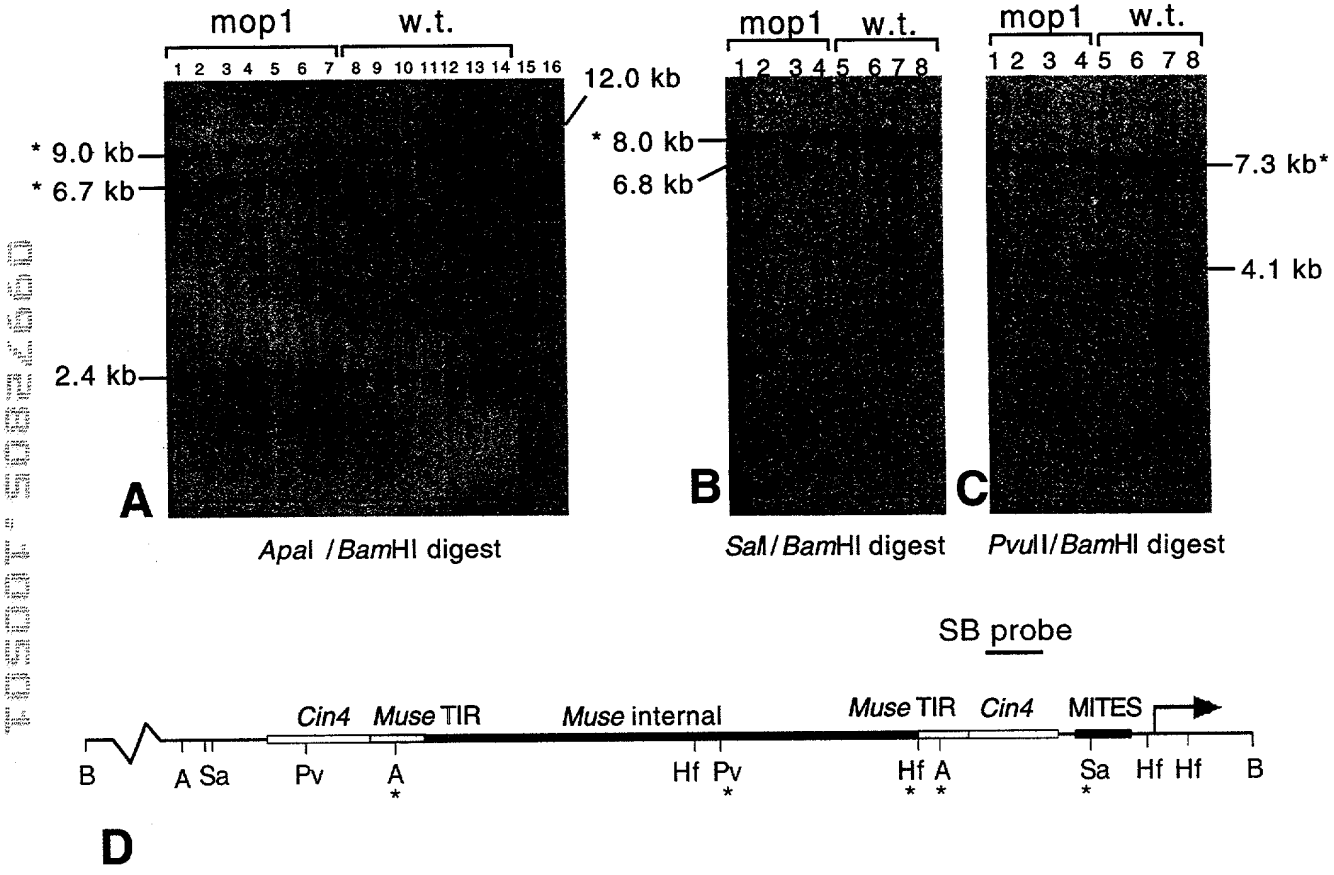

mop1
w.t.
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16
12.0 kb
* 9.0 kb
* 6.7 kb
2.4 kb
A
ApaI /BamHI digest
mop1
w.t.
1 2 3 4 5 6 7 8
* 8.0 kb
6.8 kb
B
SalI/BamHI digest
mop1
w.t.
1 2 3 4 5 6 7 8
7.3 kb*
4.1 kb
C
PvuII/BamHI digest
SB probe
Cin4
Muse TIR
Muse internal
Muse TIR
Cin4
MITES
B
A Sa
Pv
A
Hf Pv
Hf A
Sa
Hf Hf
B
D

Figure 16

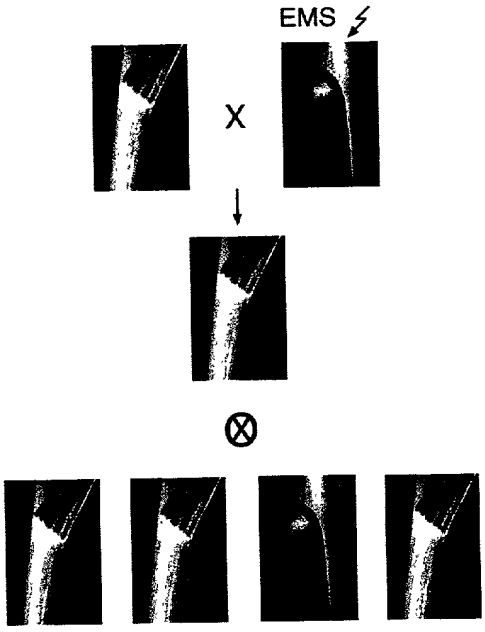


Figure 17

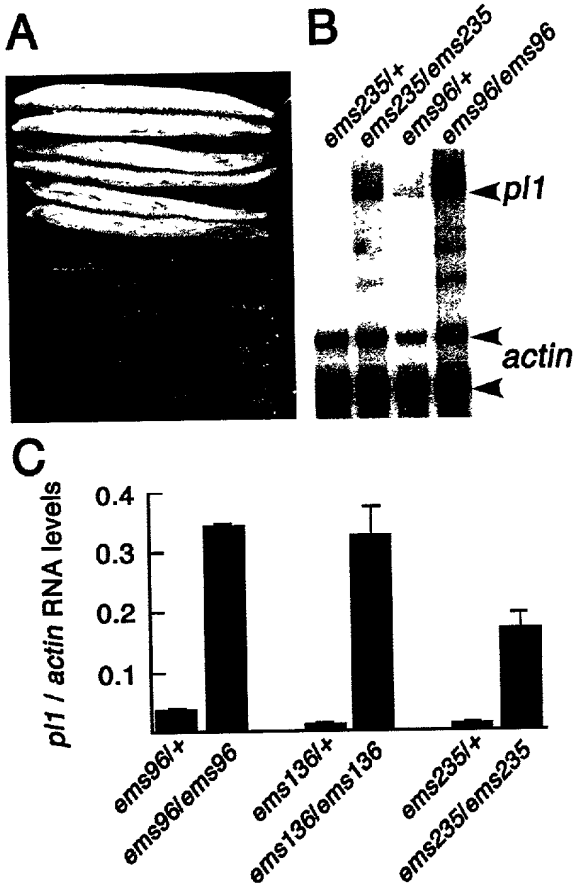

A
B
ems235/+
ems235/ems235
ems96/+
ems96/ems96
pl1
actin
C
pl1 / actin RNA levels
0.4
0.3
0.2
0.1
ems96/+
ems96/ems96
ems136/+
ems136/ems136
ems235/+
ems235/ems235

Figure 18

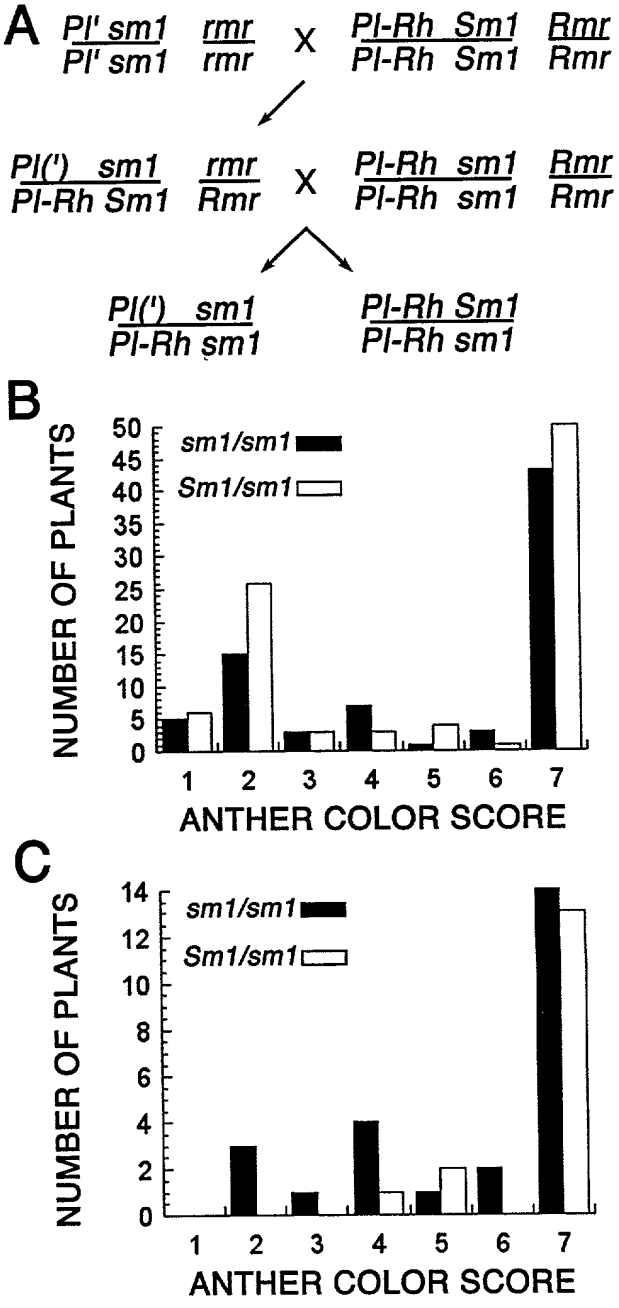

A
Pl' sm1 / Pl' sm1
rmr / rmr
X
Pl-Rh Sm1 / Pl-Rh Sm1
Rmr / Rmr
Pl(') sm1 / Pl-Rh Sm1
rmr / Rmr
X
Pl-Rh sm1 / Pl-Rh sm1
Rmr / Rmr
Pl(') sm1 / Pl-Rh sm1
Pl-Rh Sm1 / Pl-Rh sm1
B
NUMBER OF PLANTS
sm1/sm1
Sm1/sm1
ANTHER COLOR SCORE
C
NUMBER OF PLANTS
sm1/sm1
Sm1/sm1
ANTHER COLOR SCORE

Figure 19

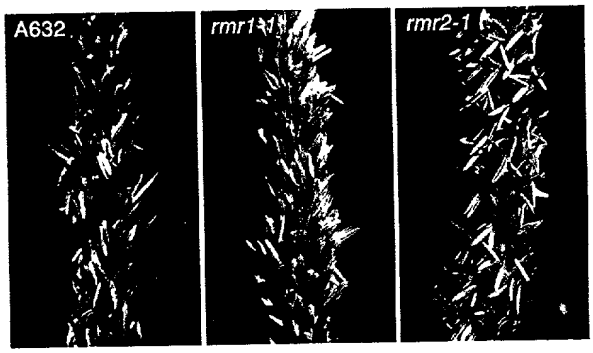


Figure 20

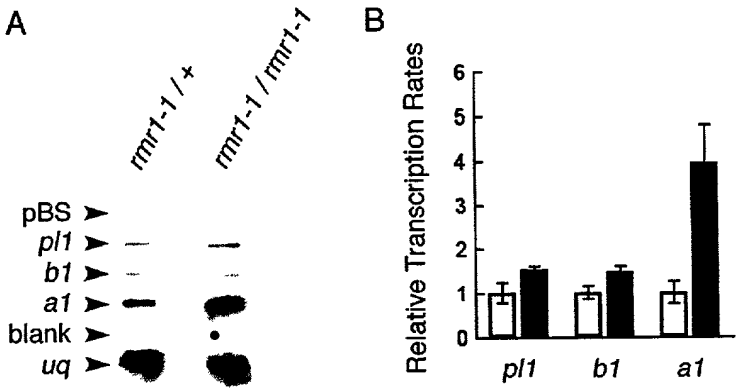

A
rmr1-1 / +
rmr1-1 / rmr1-1
pBS
pl1
b1
a1
blank
uq
B
Relative Transcription Rates
0
1
2
3
4
5
6
pl1
b1
a1

Figure 21

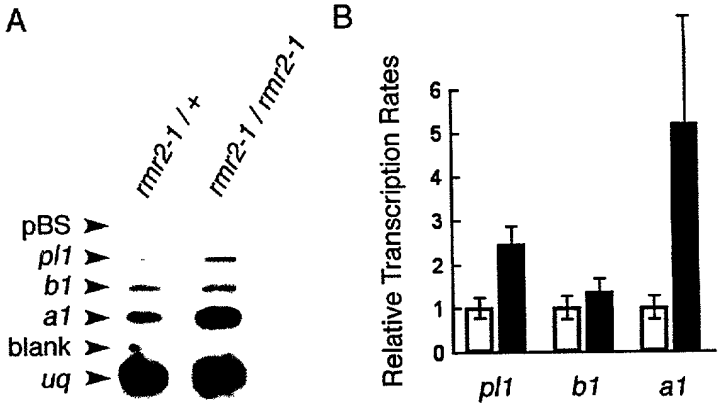

A
rmr2-1 / +
rmr2-1 / rmr2-1
pBS
pl1
b1
a1
blank
uq
B
Relative Transcription Rates
0
1
2
3
4
5
6
pl1
b1
a1

Figure 22

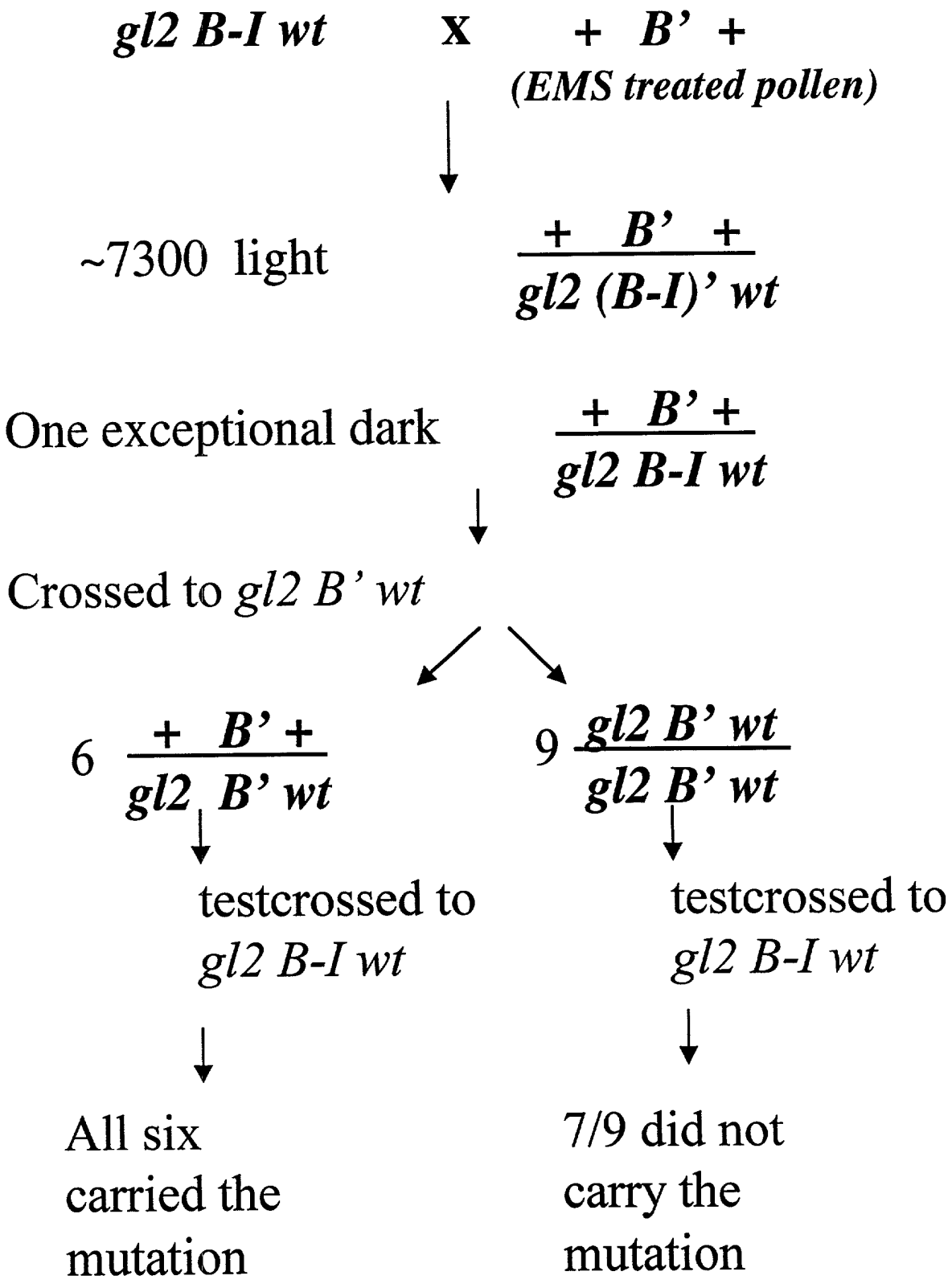

gl2 B-I wt
x
+ B' +
(EMS treated pollen)
~7300 light
+ B' +
gl2 (B-I)' wt
One exceptional dark
+ B' +
gl2 B-I wt
Crossed to gl2 B' wt
6
+ B' +
gl2 B' wt
testcrossed to
gl2 B-I wt
All six
carried the
mutation
9
gl2 B' wt
gl2 B' wt
testcrossed to
gl2 B-I wt
7/9 did not
carry the
mutation

Figure 23

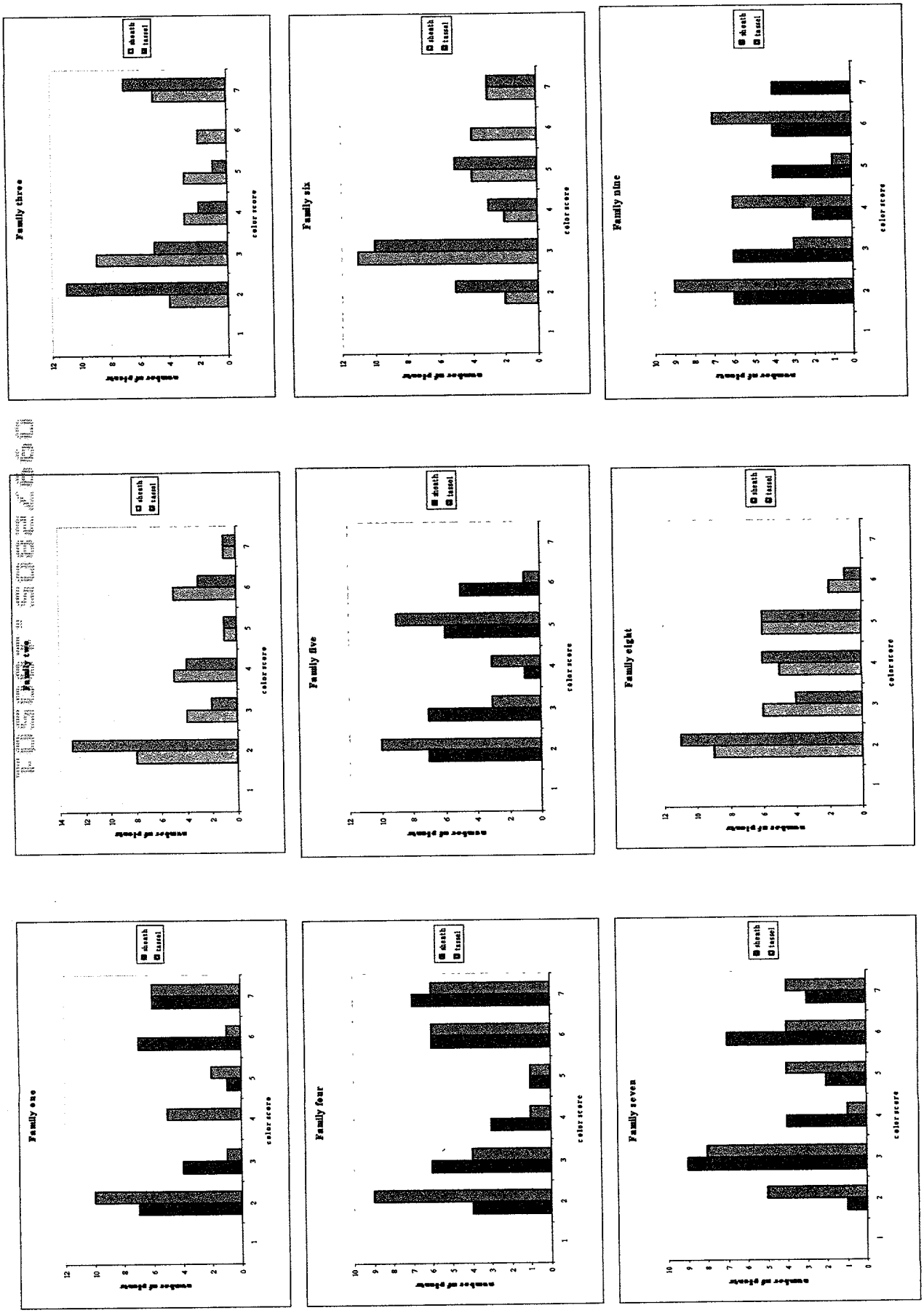

Family one
Family two
Family three
Family four
Family five
Family six
Family seven
Family eight
Family nine
sheath
tassel
color score
number of plants

Figure 24

Figure 25

EtBr stained gel
b1 RNA
A
Mop2-1/+
Mop2-1/Mop2-1
Mop2-1/Mop2-1
B
mop3-1/mop3-1
mop3-1/+
mop1-1/mop1-1
mop1-1/+
C
CC2343 light
CC2343 light
CC2343 dark
CC2343 dark

Figure 26

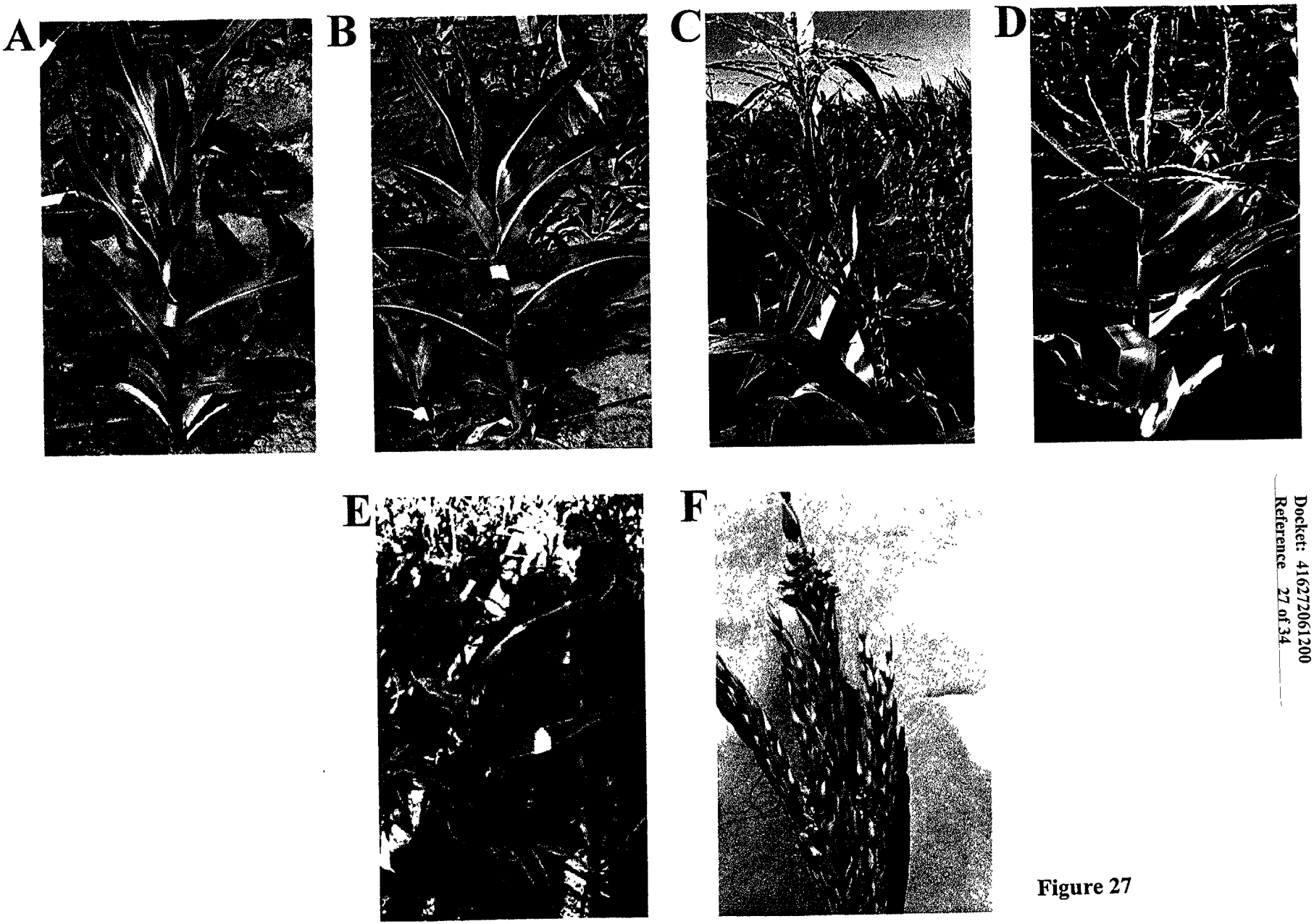


Figure 27

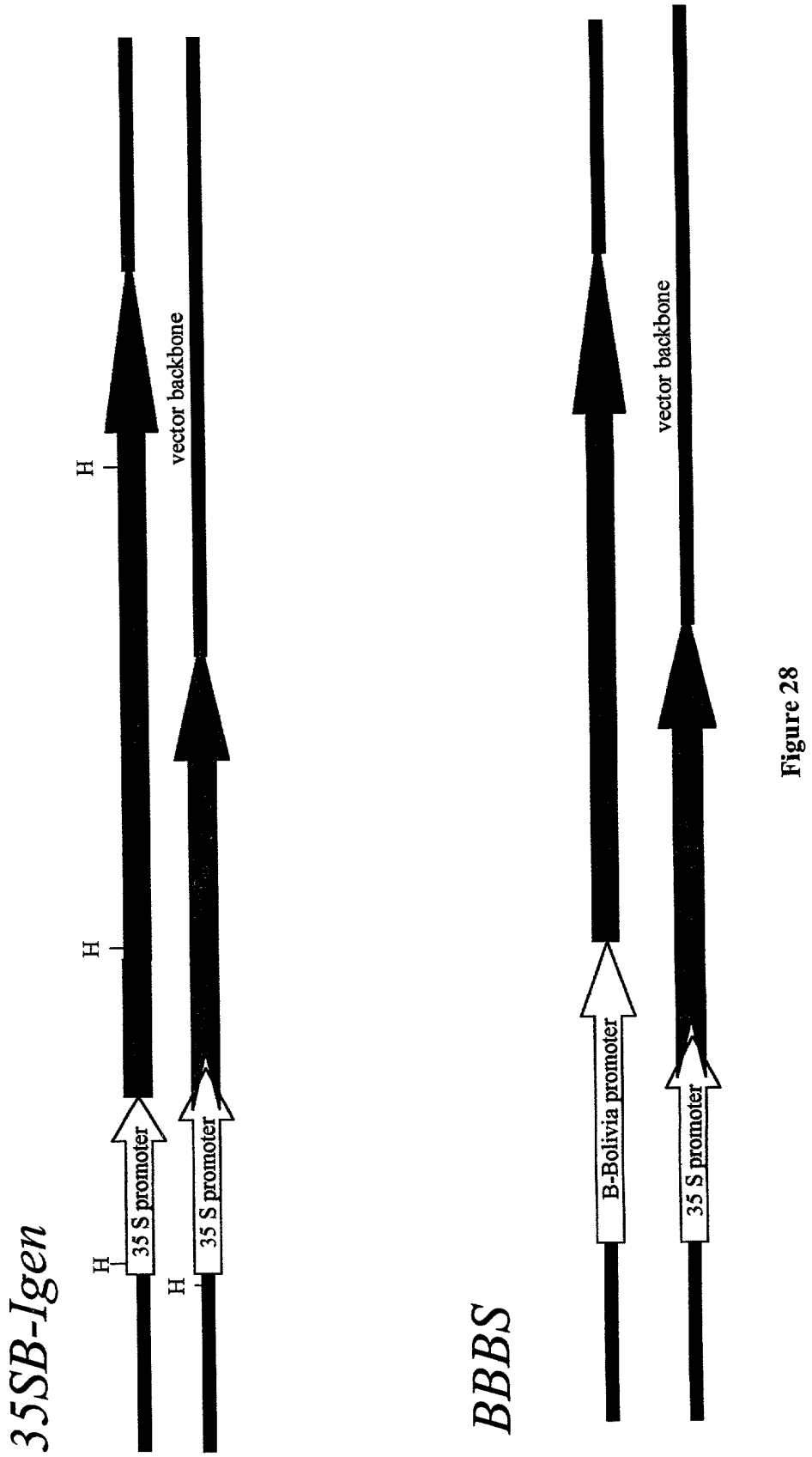

35SB-Igen
H
H
H
35 S promoter
35 S promoter
vector backbone
BBBS
B-Bolivia promoter
35 S promoter
vector backbone

Figure 28

A

B' Mop2-1/mop2 B' Mop2-1/Mop2-1

35SB-I

B

rmr2-1/rmr2-1 Rmr2/rmr2-1

35SB-I

C

B' mop1-1/mop1-1 B' Mop1/mop1-1

35SB-I

D

rmr1-1/rmr1-1 Rmr1/rmr1-1

35SB-I

E

Rmr2/rmr2-1 rmr2-1/rmr2-1

BBBS

Figure 29

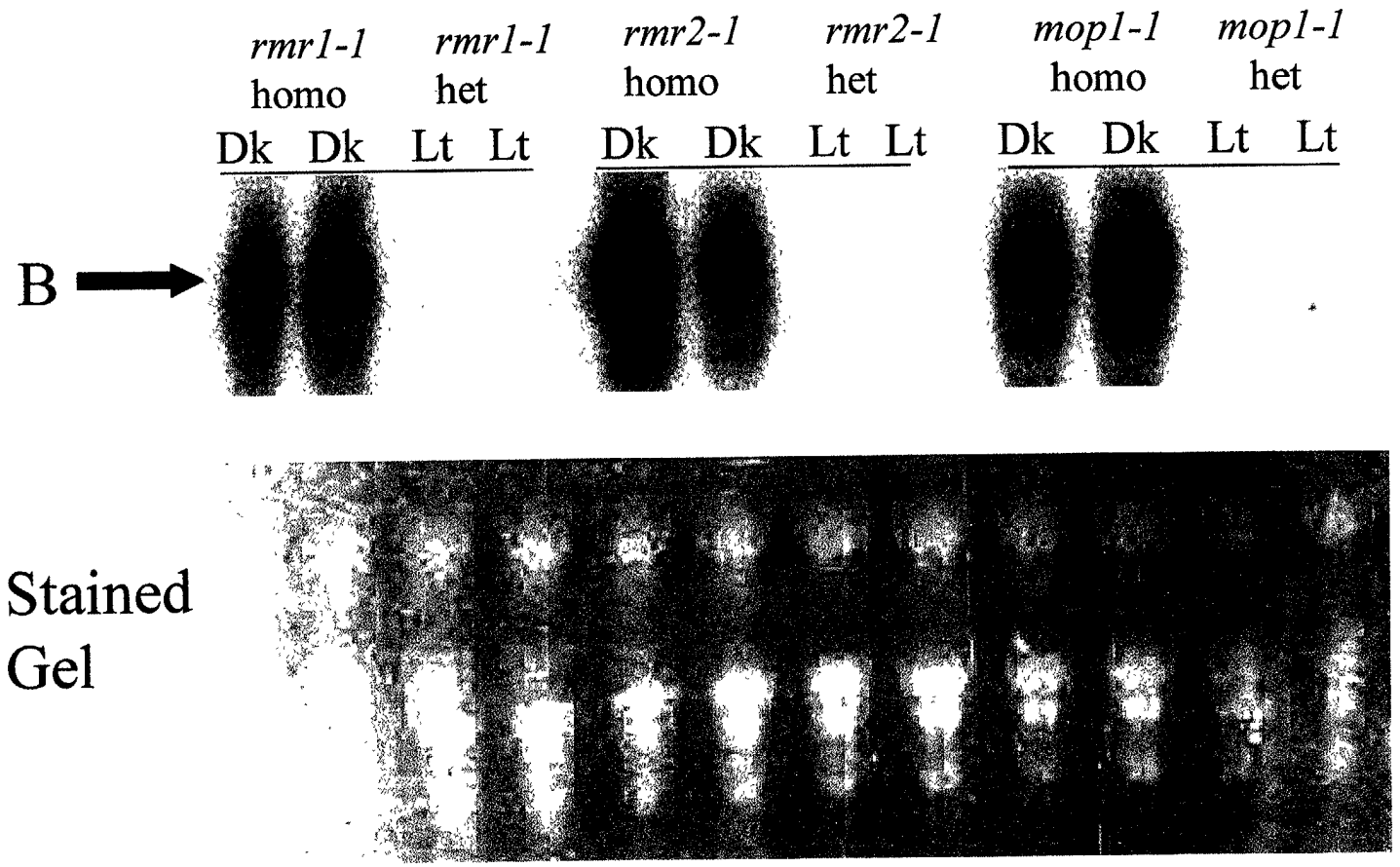


Figure 30

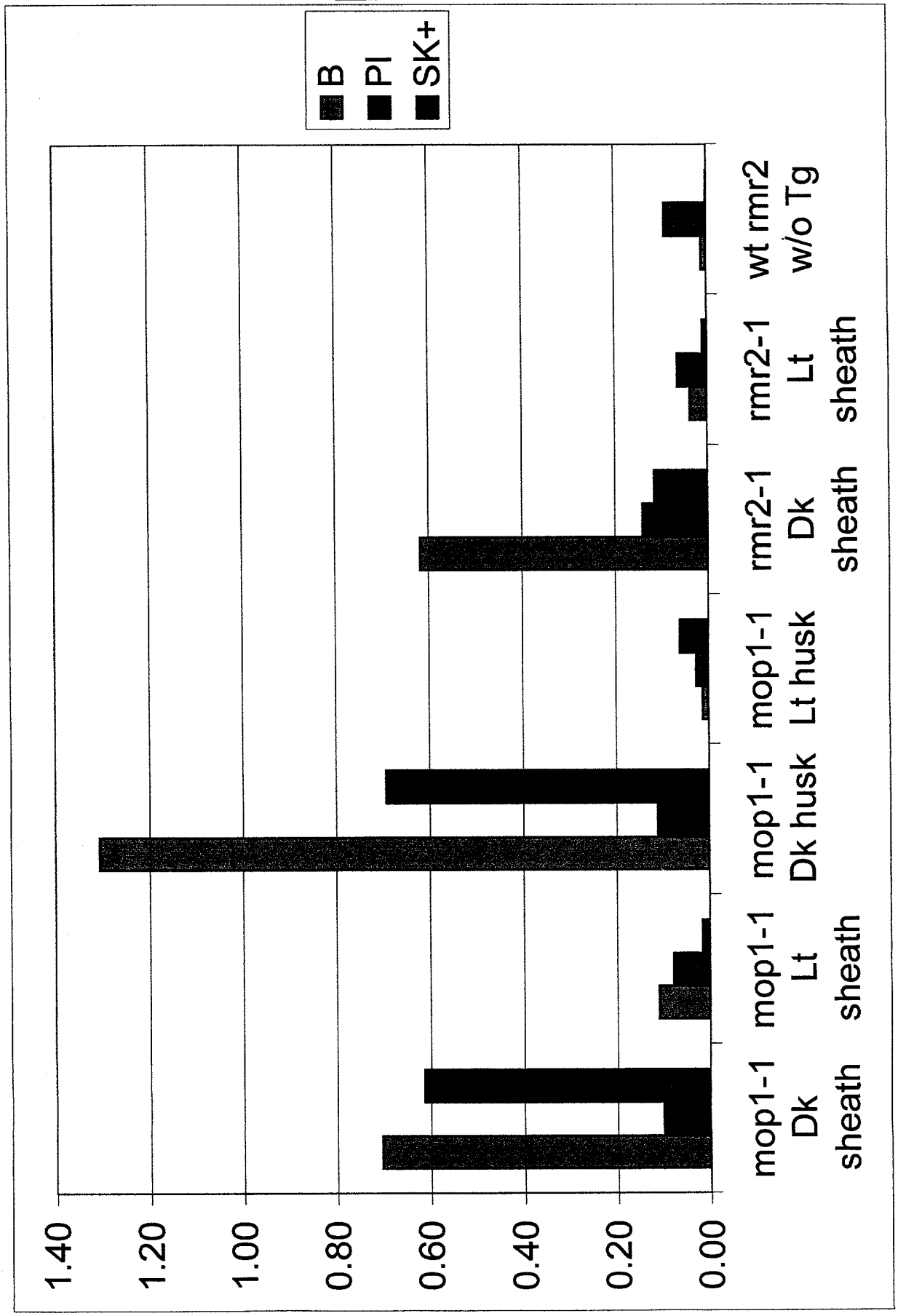

B
PI
SK+
1.40
1.20
1.00
0.80
0.60
0.40
0.20
0.00
mop1-1 Dk sheath
mop1-1 Lt sheath
mop1-1 Dk husk
mop1-1 Lt husk
rmr2-1 Dk sheath
rmr2-1 Lt sheath
wt rmr2 w/o Tg

Figure 31

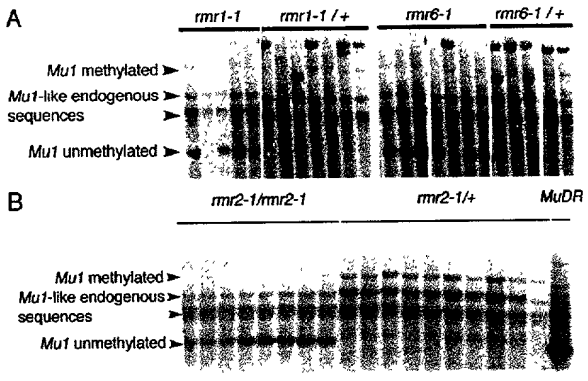

A
rmr1-1
rmr1-1 /+
rmr6-1
rmr6-1 /+
Mu1 methylated
Mu1-like endogenous sequences
Mu1 unmethylated
B
rmr2-1/rmr2-1
rmr2-1/+
MuDR
Mu1 methylated
Mu1-like endogenous sequences
Mu1 unmethylated

Figure 32

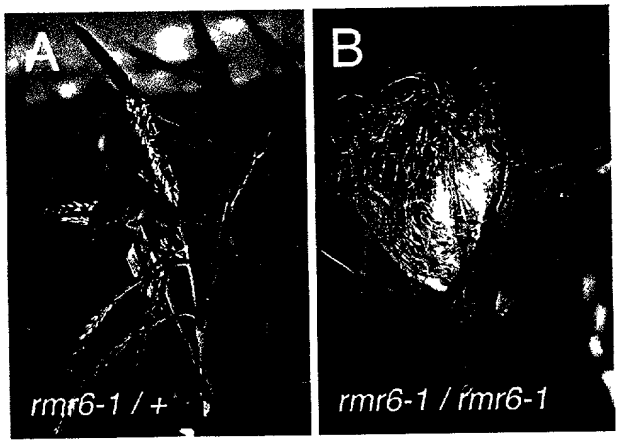


Figure 33

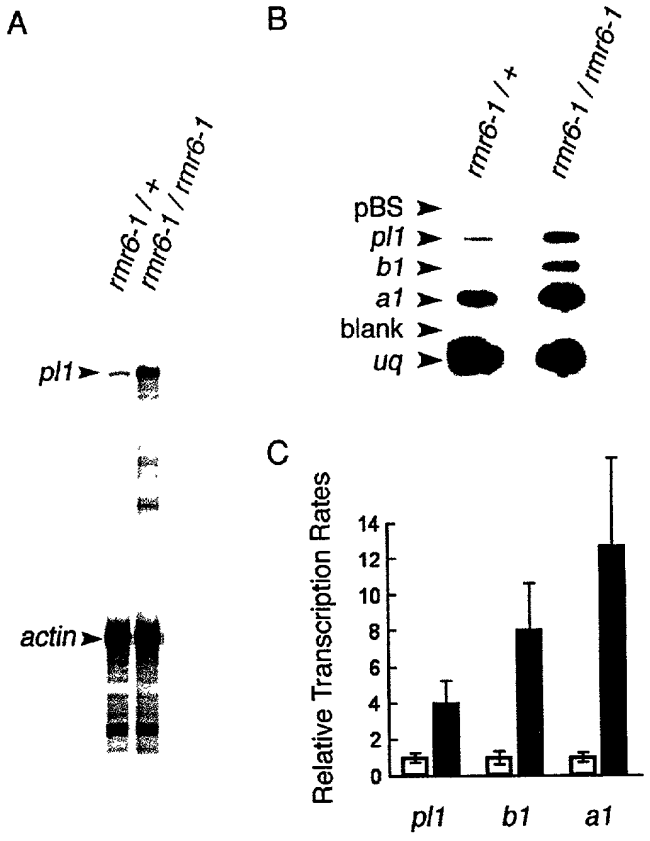

A
rmr6-1 / +
rmr6-1 / rmr6-1
pl1
actin
B
rmr6-1 / +
rmr6-1 / rmr6-1
pBS
pl1
b1
a1
blank
uq
C
Relative Transcription Rates
14
12
10
8
6
4
2
0
pl1
b1
a1

Figure 34